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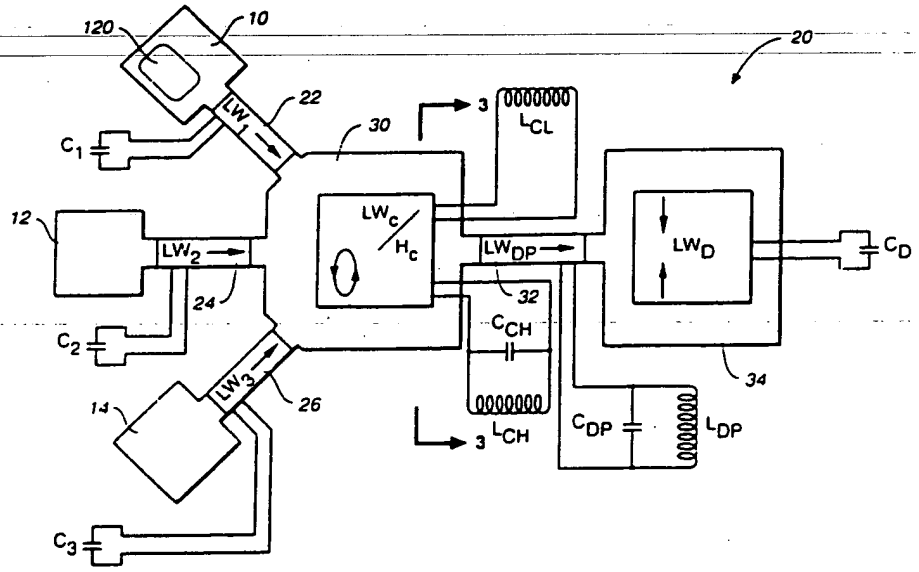

(57) Abstract

An integrated microfabricated instrument (20) for manipulation, reaction and detection of microliter to picoliter samples. The instrument (20) is suited for biochemical reactions, particularly DNA-based reactions such as the polymerase chain reaction, that require thermal cyclind since the inherently small size of the instrument facilitates rapid cycle times. The integrated nature of the instrument (20) provides accurate, contamination-free processing. The instrument (20) may include reagent reservoirs (10, 12, 14), agitators and mixers (LWc), heaters (Hc), and optical or electromechanical sensors (LWd). Ultrasonic Lamb-wave devices may be used as sensors (LWd), pumps (LW1, LW2, LW3), and agitatos (LWc).

MICROFABRICATED REACTOR

RELATED APPLICATIONS

This application is related to U.S. Patent 5,129,261, Serial No. 07/467,412, filed January 18, 1990 and application Serial No. 07/162,193, filed February 29, 1988, now abandoned, for a Plate-mode Ultrasonic Sensor. The entire disclosures of these applications are hereby incorporated by reference.

BACKGROUND OF THE INVENTION

The present invention relates generally to instruments for chemical reaction control, product and reactant manipulations, detection of participating reactants and resultant products, and more particularly to integrated microfabricated instruments which perform microscale chemical reactions involving precise control of parameters of the reactions. The parameters of the reaction controlled by the instrument may be temperature, pressure, concentration of reactants, the intensity or frequency of incident light, electromagnetic fields, or ultrasonic pressure waves, etc.

The term "integrated microfabrication" is used herein to refer to all processes used for batch production of semiconductor microelectronics, and all related microfabrication processes such as LIGA (see R. S. Muller, R. T. Howe, S. D. Senturia, R. L. Smith, and R. M. White, ed. MICROSENSORS, IEEE Press, 472 pages, 1990). Microfabrication technologies include, but are not limited to, sputtering, electrodeposition, low-pressure vapor deposition, photolithography and etching. Microfabricated devices are usually formed on crystalline semiconductor substrates such as silicon or gallium arsenide. Noncrystalline materials such as glass or certain polymers may be used although crystalline materials provide certain advantages. The shapes of crystalline devices can be precisely controlled since etched surfaces are generally crystal planes, and crystalline materials may be bonded by processes such as fusion at elevated temperatures or the field-assisted method (Mallory bonding). Materials which are not semiconductors, such as quartz or glass, may be used, though semiconductor materials provide the advantage that electronic circuitry may be integrated into the system by the use of conventional integrated-circuit fabrication techniques.

Monolithic microfabrication technology now allows the production of electrical, mechanical, electromechanical, optical, chemical and thermal devices including pumps, valves, heaters, mixers and species detectors for microliter to nanoliter quantities of solids, liquids and gases. Microscale sensors include optical waveguide probes and ultrasonic flexural-wave sensors. The integration of these devices into a single system allows for the batch production of microscale reactor-based analytical instruments. Integrated microinstruments

may be applied to biochemical, inorganic, or organic chemical reactions to perform biomedical and environmental diagnostics, and biotechnological processing and detection.

Such integrated microfabricated devices can be manufactured in batch quantities with high precision, yet low cost, thereby making recyclable and/or disposable single-use devices practical. Alternatively, the instrument may consist of an array of reaction instruments which are to operate in parallel to simultaneously perform a number of related reactions. Operation of such instruments is easily automated, further reducing costs. Since the analysis can be performed *in situ*, the likelihood of contamination is very low. Because of the inherently small sizes of such devices, the heating and cooling can be extremely rapid, and the devices can have very low power requirements. Such devices may be powered by batteries or by electromagnetic, capacitive, inductive or optical coupling.

Small volumes and high surface-area to volume ratios provide microfabricated reaction instruments with a high level of control of the parameters of a reaction. Heaters may produce temperature cycling or ramping, sonochemical and sonophysical changes in conformational structures may be produced by ultrasound transducers, and polymerizations may be generated by incident optical radiation.

Synthesis reactions, and especially synthesis chain reactions such as the polymerase chain reaction (PCR), are particularly well-suited for microfabricated reaction instruments. PCR can selectively amplify a single molecule of DNA (or RNA)

lower temperature, and maintaining the temperature at that lower temperature. The rate at which the sample is heated is generally limited by the heater rather than the rate of heat transfer to the sample. Presently, each of the four stages of a thermal cycle requires approximately one minute, and the time required for twenty to forty complete thermal cycles is therefore from about one to three hours. The cycling time has been reduced by performing the PCR reaction in capillary tubes (see C. T. Wittwer, G. C. Fillmore, and D. J. Garling, Analytical Biochemistry, 186, pp. 328-331 (1990)). A high-power forced air heater was used to heat the tubes. The thinnest capillary tubes contained a sample volume of about ten microliters. Each cycle consisted of a heating step, a waiting period, a cooling step and another waiting period, and each step required approximately fifteen seconds.

Although the PCR reaction requires thermal cycling of the reagents, any reaction that benefits from precise temperature control, and/or rapid thermal cycling, thermal ramping, or any other temperature variation of reagents with time (hereinafter to be referred to as temperature programming) will be well suited for the microfabricated reaction instrument of the present invention.

An object of the present invention is therefore to provide a integrated microfabricated reactor.

Another object of the present invention is to provide a reactor-based instrument for inorganic, organic and biochemical reactions, and in particular for diagnostics.